

PRELIMINARY AMENDMENT  
Continuation of U.S. Appln. No. 09/125,944

REMARKS

Claims 1-18 have been amended to remove improper multiple dependencies and for editorial purposes only. No new matter has been added.

Entry and consideration of this Amendment is respectfully requested.

Respectfully submitted,



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PRELIMINARY AMENDMENT  
Continuation of U.S. Appln. No. 09/125,944

APPENDIX

VERSION WITH MARKINGS TO SHOW CHANGES MADE

IN THE CLAIMS:

**The claims are amended as follows:**

1. (amended) A method for measuring protease which comprises the steps of:
  - (1) contacting ~~bringing~~ a sample containing protease ~~into contact~~ with a thin membrane which comprises a protease substrate together with a hardening agent ~~and is formed~~ on a surface of a support; and
  - (2) detecting ~~the~~ a trace of digestion formed on the thin membrane by the action of protease.
2. (amended) A method for measuring protease which comprises the steps of:
  - (1) contacting ~~bringing~~ one of two substantially continuous slices of a biological sample ~~into contact~~ with a thin membrane which comprises a protease substrate together with a hardening agent ~~and is formed~~ on a surface of a support;
  - (2) detecting ~~the~~ a trace of digestion formed on the thin membrane by the action of protease; and
  - (3) comparing the trace of digestion with a histopathological preparation prepared from the other slice.
3. (amended) A method for measuring protease which comprises the steps of:
  - (1) contacting ~~bringing~~ one of two or more substantially continuous slices of a biological sample ~~into contact~~ with a thin membrane which comprises a protease substrate together with a hardening agent ~~and is formed~~ on a surface of a support;

PRELIMINARY AMENDMENT  
Continuation of U.S. Appln. No. 09/125,944

(2) contacting bringing the remaining slices ~~into contact~~ with a thin membrane which comprises a protease substrate, a hardening agent, and a protease inhibitor ~~and is formed on a~~ surface of a support;

(3) detecting traces of digestion formed on the thin membranes by the action of protease; and

(4) comparing the trace of digestion on the thin membrane used in ~~the~~ step (1) with the trace of digestion on the thin membrane used in ~~the~~ step (2).

4. (amended) A method for measuring protease which comprises the steps of:

(1) contacting bringing one of two or more substantially continuous slices of a biological sample ~~into contact~~ with a thin membrane which comprises a protease substrate together with a hardening agent ~~and is formed on a surface of a support~~;

(2) contacting bringing the remaining slices ~~into contact~~ with a thin membrane which comprises a protease substrate different from the protease substrate ~~contained~~ present in the thin membrane used in ~~the~~ step (1) together with a hardening agent ~~and is formed on a surface of a support~~;

(3) detecting traces of digestion formed on the thin membranes by the action of protease; and

(4) comparing the trace of digestion on the thin membrane used in ~~the~~ step (1) with the trace of digestion on the thin membrane used in ~~the~~ step (2).

5. (amended) A method for measuring protease which comprises the steps of:

(1) contacting bringing a sample containing protease ~~into contact~~ with a thin membrane which comprises at least the following two layers: layer (a) which contains a protease

PRELIMINARY AMENDMENT

Continuation of U.S. Appln. No. 09/125,944

substrate, a hardening agent, and a protease inhibitor ~~and is~~ formed on a surface of a support, and layer (b) which contains a protease substrate and a hardening agent ~~and is~~ laminated on ~~the~~ layer (a);

(2) detecting traces of digestion formed on the thin membrane by the action of protease; and

(3) comparing the trace of digestion on ~~the~~ layer (a) with the trace of digestion on ~~the~~ layer (b).

6. (amended) A method for measuring protease which comprises the steps of:

(1) contacting ~~bringing~~ a sample containing protease ~~into contact~~ with a thin membrane which comprises at least the following two layers: layer (a) which contains a protease substrate together with a hardening agent ~~and is~~ formed on a surface of a support, and layer (b) which contains a protease substrate different from the protease substrate present ~~contained~~ in the layer (a) together with a hardening agent ~~and is~~ laminated on ~~the~~ layer (a);

(2) detecting traces of digestion formed on the thin membrane by the action of protease; and

(3) comparing the trace of digestion on ~~the~~ layer (a) with the trace of digestion on ~~the~~ layer (b).

7. (amended) The method of claim 1 ~~any one of claims 1 to 6~~ wherein the protease substrate is selected from the group consisting of collagen, gelatin, proteoglycan, fibronectin, laminin, elastin, and casein.

8. (amended) The method of claim 1 ~~any one of claims 1 to 7~~ wherein the sample is a biological sample isolated or collected from a patient.

PRELIMINARY AMENDMENT  
Continuation of U.S. Appln. No. 09/125,944

9. (amended) The method of claim 1 ~~any one of claims 1 to 8~~ wherein the detecting ~~detection is performed~~ by using a thin membrane containing one or more substances selected from the group consisting of metals, metal oxides, pigments and dyes and having a maximum transmission density of 0.01 or higher at a wavelength ranging from 400 nm to 700 nm.

10. (amended) The method of claim 1 ~~any one of claims 1 to 9~~ wherein the protease is a matrix metalloproteinase.

11. (amended) A thin membrane for measuring protease which contains a protease substrate together with a hardening agent ~~and is~~ formed on a surface of a support.

12. (amended) The thin membrane of claim 11 which comprises at least the following two layers:

layer (a) which comprises a protease substrate, a hardening agent and a protease inhibitor ~~and is~~ formed on a surface of a support, and layer (b) which contains a protease substrate together with a hardening agent ~~and is~~ laminated on ~~the~~ layer (a).

13. (amended) The thin membrane of claim 11 which comprises at least the following two layers: layer (a) which comprises a protease substrate together with a hardening agent ~~and is~~ formed on a surface of a support, and layer (b) which comprises a protease substrate different from the protease substrate ~~contained present in~~ ~~the~~ layer (a) together with a hardening agent ~~and is~~ laminated on ~~the~~ layer (a).

14. (amended) The thin membrane of claim 11 ~~any one of claims 11 to 13~~ which ~~comprise~~ comprises one or more substances selected from the group consisting of metals, metal oxides, pigments and dyes and have a maximum transmission density of 0.01 or higher at a wavelength ranging from 400 nm to 700 nm.

PRELIMINARY AMENDMENT

Continuation of U.S. Appln. No. 09/125,944

15. (amended) The thin membrane of claim 11 ~~any one of claims 11 to 14~~ wherein the support is ~~selected from~~ a microscope slide ~~and~~ or a polyethylene terephthalate film.

16. (amended) The thin membrane of claim 11 ~~any one of claims 11 to 15~~ wherein an undercoat layer is ~~present provided~~ between the support and the thin membrane.

17. (amended) A method of diagnosing a disease involving protease which comprises the steps of:

(1) ~~contacting bringing~~ a biological sample isolated or collected from a patient ~~into~~ ~~contact~~ with a thin membrane which comprises a protease substrate together with a hardening agent ~~and is~~ formed on a surface of a support; and

(2) detecting the trace of digestion formed on the thin membrane by the action of protease.

18. (amended) The method of claim 17 wherein the disease is selected from the group consisting of cancer, rheumatic diseases, periodontal diseases and alveolar pyorrhea.